

NOTEWORTHY COLLECTION

CALIFORNIA

GRACILARIOPSIS CHORDA (Holmes) Ohmi 1958:24 (GRACILARIACEAE).—Monterey Co., attached to a small 5 × 12 m concrete boat launch ramp in the lower intertidal at Kirby Park, Elkhorn Slough, 36°50'23.53"N, 121°44'37"W, sterile and cystocarpic, 25 October 2011, *J. R. Hughey s.n.* (UC 1997139); 15 November 2011, *J. R. Hughey s.n.* (UC 1997140, UC 1997141, UC 1997142), and 7 May 2012, *J. R. Hughey s.n.* (UC 1997143, UC 1997144, UC 1997145, UC 1997146).

Previous knowledge. *Gracilariopsis chorda* (Holmes) Ohmi is a marine red alga that is native to Japan, China, and Korea (Holmes 1896; Kim et al. 2008). The type collection of this species, from Enoura, Shizuoka Prefecture, Japan, was characterized as having a large (up to 1 m), succulent, purple-colored thallus with only three branches off the main axis (Holmes 1896). *Gracilariopsis chorda* was originally assigned by Holmes (1896) to *Gracilaria*, but it was later transferred by Ohmi (1958) to *Gracilariopsis* because it lacked nutritive filaments that extend into the pericarp, one of the defining features of *Gracilaria* (Fredericq and Hommersand 1989). *Gracilariopsis chorda* has coarse, reddish-brown thalli that extend up to 110 cm in length with cylindrical 2–3 mm, sparingly to profusely branched axes and filiform branchlets (Ohmi 1958; Yamamoto 1978; Kim et al. 2008; M. S. Kim et al. 2010). The medullary cells of *G. chorda* are large (612–700 µm) and appear almost empty (Ohmi 1958), with an abrupt transition in cell size from the cortex to the medulla (Kim et al. 2008). On the basis of an analysis of *cox1* and *rbcL* gene sequences from 22 specimens from Asia, Kim et al. (2008) agreed with Ohmi's placement of *G. chorda* in *Gracilariopsis* at the rank of species.

Significance. First report of *G. chorda* in the northeastern Pacific. The specimens of *G. chorda* collected from Elkhorn Slough are in agreement with published illustrations and descriptions of this species. Anatomical examination of this alga show the cortex to be abbreviated and distinct from the medullary cells, and the medulla to consist of large globular cells (~500 µm in diameter). The cystocarps of *G. chorda* are conspicuous and restricted to the middle thallus. *Gracilariopsis chorda* is red in color when fresh (drying to dark brown or black), while the native species, *Gracilariopsis andersonii* (Grunow) E.Y. Dawson, is straw-colored (Abbott and Hollenberg 1976), and the recently reported invasive seaweed *Gracilaria vermiculophylla* (Ohmi) Papenfuss is dark brown (S. Y. Kim et al. 2010). *Gracilaria vermiculophylla* does not occur on the ramp at the launch facility, but is abundant immediately adjacent to the ramp where it grows on small pebbles and shells partly buried in the mud. It is common throughout the slough. *Gracilariopsis andersonii* is found only near the mouth. Analysis of the *rbcL* gene of *G. chorda* from Elkhorn Slough yielded a DNA sequence (GenBank JX262420) that was identical to eight sequences from South Korea, four from Japan, one from China, and one from an introduced population in the Gulf of Morbihan, Brittany, France (Mineur et al. GenBank, unpublished). Of the five *rbcL*

haplotypes reported by S. Y. Kim et al. (2010) for *G. chorda*, the Elkhorn population is assigned to R2, the largest of the haplogroups.

GRATELOUPIA ASIATICA Kawaguchi & Wang 2001: 435 (HALYMENTIACEAE).—San Diego Co., collected from the dock below the Coronado Boathouse Restaurant, Coronado Island, San Diego, 32°40'46.81"N, 117°10'29.30"W, tetrasporangial, 25 June 2012, *J. R. Hughey s.n.* (UC 1997160).

Previous knowledge. *Grateloupia asiatica* is native to Japan, China, and Korea (Kawaguchi et al. 2001; De Clerck et al. 2005; Lee et al. 2009) (type locality: Tsuyazaki, Fukuoka Prefecture, northern Kyushu, Sea of Japan). It is characterized as having dark red, 10–15 cm high thalli with pinnate branching (Kawaguchi et al. 2001). Although this species has been known in Asia as *G. filicina* (J.V. Lamouroux) C. Agardh (Kawaguchi et al. 2001), morphological and molecular analyses have demonstrated that *G. filicina* is restricted to the Mediterranean basin (Kawaguchi et al. 2001; De Clerck et al. 2005). *Grateloupia asiatica* is said to differ from *G. filicina* by its habit and texture (thin and soft with wider axes), vegetative anatomy (denser medulla), and reproductive structures (scattered and with an large, oval auxiliary cell). *Grateloupia asiatica* and other *Grateloupia* species were recently reported as introduced to Thau Lagoon, Mediterranean, France (Verlaque et al. 2005).

Significance. This is the first report of *G. asiatica* in the eastern Pacific ocean. The single specimen collected at Coronado strongly resembles the illustration of *G. filicina* in Abbott and Hollenberg (1976, fig. 384), a species reportedly rare in California, but occurring on Santa Catalina Island. The morphology and shape of the tetraspores of the Coronado specimen also shows similarities to *G. asiatica* (Kawaguchi et al. 2001; Verlaque et al. 2005). Identification of the Coronado specimen as *G. asiatica* was confirmed with an *rbcL* DNA sequence (GenBank JX307635). The sequence differed by only one bp from two sequences of *G. asiatica* from Fukuoka, Noshima; two bp from Fukuoka, Hakata Bay; three bp from Kochi, Sukumo, Minatoura, Japan (Kawaguchi et al. 2001); and seven bp from two specimens from Qingdao, Shandong Province, China (De Clerck et al. 2005). The DNA sequence of the Coronado specimen did not match the nonnative population (i.e., *G. filicina*) in Thau lagoon, differing by three bp (Verlaque et al. 2005). All of these representatives fit comfortably within reported intra-specific sequence divergences (<1% = 0 to 14 bp) for *rbcL* gene sequences in red algae (Freshwater and Rueness 1994). The vector for the Coronado introduction is likely hull fouling, because San Diego Bay is home to a large number of pleasure, commercial, and naval vessels. The appearance of *G. asiatica* was predicted by Miller et al. (2011) based on introduction patterns into the Californias by other nonnative seaweeds. Since the range of *G. filicina* is restricted to the Mediterranean, and *G. asiatica* strongly resembles *G. filicina*, it is probable that previous collections identified as *G. filicina* from southern California are assignable to *G. asiatica*. A genetic study of both recent and historical specimens from California will be

required to assess the distribution of *G. asiatica* in the eastern Pacific Ocean.

ULVA CLATHRATIOIDES L. G. Kraft, Kraft & R. F. Waller 2010:1273 (ULVACEAE).—Monterey Co., covering mud in the upper intertidal at railroad bridge, Elkhorn Slough, 36°51'27.25"N, 121°45'20.75"W, fertile, 29 May 2012, *J. R. Hughey s.n.* (UC 1997147); floating and attached to shells in the upper intertidal at Kirby Park, Elkhorn Slough, 36°50'23.53"N, 121°44'37"W, fertile, 7 May 2012, *J. R. Hughey s.n.* (UC 1997148), and 29 May 2012, *J. R. Hughey s.n.* (UC 1997149); attached to boat launch dock near the marina docks at Elkhorn Slough, 36°48'46"N, 121°47'14"W, fertile, 29 May 2012, *J. R. Hughey s.n.* (UC 1997150, UC 1997151).

Previous knowledge. *Ulva clathratioides* was described from southern Australia where it grows at 20–30 cm depth at low tide and on rocks on inner-reef flats (type locality: Point Lonsdale, Victoria [Kraft et al. 2010]). It forms small (3–5 cm), dense tufts, that are highly irregular in branching, and typically proliferous. The chloroplasts in this species occupy a narrow, peripheral position in cells. In section, the lateral branches contain cells that are embedded in a thick, extracellular matrix that extends up to 45 µm into the lumen of the branch (Kraft et al. 2010). On the basis of these features, *U. clathratioides* was originally and tentatively identified as *U. clathrata* (Roth) Greville by the authors. However, an analysis of nuclear (internal transcribed spacer) and plastid (*rbcL*) DNA sequences from two specimens showed it to be unrelated to the European *U. clathrata*.

Significance. First report of *U. clathratioides* in the eastern Pacific. Mature thalli are large (up to 35 cm high), pale to dark green in color, and appear hairy. Axes bear many multiseriate branches that vary in length from 200 µm to 1.5 cm. In young plants and branches, the cells are arranged in longitudinal rows, but become disordered as they mature. The chloroplasts are as described by Kraft et al. (2010), however the firm, hyaline matrix reportedly characteristic of this species was not observed in any thalli from Elkhorn Slough. The *rbcL* gene from four specimens from Elkhorn Slough (one near the head, two in the central region, and one at the mouth) was analyzed; the sequences (1171 bp) were identical for all four (GenBank JX262426–JX262429). Comparison of the Elkhorn Slough sequences to those in GenBank showed an exact match to a specimen labeled *Ulva* sp. 2 from North Island, Bay of Islands, Russell, New Zealand (Heesch et al. 2007), but there was only 81% sequence coverage. The missing 19% of sequence near the 3' end of the *rbcL* gene contains two polymorphic sites that are unique to the Elkhorn Slough specimens. It is therefore not possible to assume that the Elkhorn population is identical in sequence to *Ulva* sp. 2 from New Zealand. *Ulva* sp. 2, like *U. clathratioides* in Australia, forms unbranched or branched tubular blades up to 3 cm high that grow in high intertidal tide pools on rocks, shells, or other algae (Heesch et al. 2007). It was characterized, but not named, as native to New Zealand in a technical report to the Ministry of Primary Industries (Heesch et al. 2007). Comparison against complete sequences revealed that Elkhorn specimens differed by three bp from two specimens from Point Lonsdale, Victoria, Australia (= *U. clathratioides* type locality, Kraft et al. 2010) and two specimens from Maili Point, Oahu Island, USA (O'Kelly et al. 2010). The Oahu specimens (OTU 5) are short (1–3 cm high) unbranched plants, that superficially resemble *Ulva* sp. 2. The Elkhorn Slough material also

differed by four bp from one specimen from Okinawa, Ishigaki Island, Japan (Horimoto et al. GenBank, unpublished), and ten specimens from South and North Islands, New Zealand (Heesch et al. 2009). Since interspecific sequence divergence for the *rbcL* gene in *Ulva* ranges from zero to five bp (Kraft et al. 2010), these results indicate that these specimens from New Zealand, Australia, Japan, and California belong to the same species. The status of *U. clathratioides* in California as a native or nonnative species requires further investigation. Additional field and molecular study will likely reveal that *U. clathratioides* occurs in other brackish water habitats in the eastern Pacific Ocean.

ULVA PERTUSA Kjellman 1897:4 (ULVACEAE).—Marin Co., covering the shoreline on rocks and on shells in the upper intertidal at Marshall, Tomales Bay, 38°09'42.70"N, 122°53'37.54"W, fertile, 20 July 2011, *J. R. Hughey s.n.* (UC 1997152); Monterey Co., on shells buried in the mud near boat launch ramp and boat dock in the middle intertidal at Kirby Park, Elkhorn Slough, 36°50'23.53"N, 121°44'37"W, fertile, 25 October 2011, *J. R. Hughey s.n.* (UC 1997153, UC 1997155); on rocks under foot bridge at South Marsh Loop, Elkhorn Slough, 36°49'11.64"N, 121°44'13.59"W, fertile, 3 May 2012, *J. R. Hughey s.n.* (UC 1997156); attached to boat launch facility near the marina docks at Elkhorn Slough, 36°48'46"N, 121°47'14"W, fertile, 29 May 2012, *J. R. Hughey s.n.* (UC 1997157); attached to floating dock in the Monterey Marina, Monterey Harbor, Monterey, 36°36'07"N, 121°53'25"W, fertile, 20 September 2011, *J. R. Hughey s.n.* (UC 1997158, UC 1997159).

Previous knowledge. *Ulva pertusa* is native to Asia (syntype localities: Hakodate, Yenoshima, and Yokohama, Japan (Kjellman 1897). This species is a known exotic (Verlaque et al. 2002) that occurs worldwide (Guiry and Guiry 2012). *Ulva pertusa* was recently documented from four localities in Baja California (Aguilar-Rosas et al. 2008) and three in southern California (Mission Bay, La Jolla, Newport Bay) (Hayden and Waaland 2004). The thallus is characterized as 1) lobed, perforated, and lacking tooth-like protuberances on the margins; 2) wrinkled in the basal portion; 3) of variable thickness, approximately 500 µm thick above the holdfast, 90–156 µm in the lower parts, and 34–50 µm on the margins; 4) composed of unordered, round cells with 1–3 pyrenoids; 5) forming a broad, linear reproductive rim (Verlaque et al. 2002).

Significance. This is the first report of *Ulva pertusa* in central and northern California. The specimens of *U. pertusa* collected from Tomales Bay, Elkhorn Slough, and the Monterey Marina are in good morphological agreement with the description above. *Ulva pertusa* grows intermixed with native *U. rigida* C. Agardh at the mouth of Elkhorn Slough and *U. lobata* (Kützinger) Harvey in the Monterey Marina. Compared to the native species, *U. pertusa* differs most notably in texture (the native species is fleshy and lax while *U. pertusa* is waxy and very rigid, appearing plastic) and cell shape (those of the native species are bullet-shaped to quadrate while those of *U. pertusa* are rounded). Identification of *U. pertusa* was confirmed using *rbcL* DNA sequences (GenBank JX262421–JX262425). The sequence from the Tomales Bay specimen did not match any accessions deposited in GenBank, and differed by 1 bp from the Monterey Marina and three Elkhorn Slough sequences, which were identical. The specimens from central California were the same as sequences of *U. pertusa* from La Jolla and Newport

Bay, and plants from Australia, China, Japan (excluding a few ambiguous nucleotides), New Zealand, and Spain. All of the above specimens differed by 2 bp from the plant from Mission Bay, California.

To determine if historical collections from the Monterey Marina are assignable to *U. pertusa*, four specimens resembling this species from the Gilbert M. Smith Herbarium at Hopkins Marine Station, Pacific Grove, California, were analyzed. The four plants included isotype material of *U. expansa* (*Ulva fasciata* forma *expansa* Setchell, P.B.-A LXXVII, GMS 8034, 1901) and blades identified by I. A. Abbott as *U. expansa* (GMS 7779, GMS 8025) and *U. lobata* (GMS 8043) collected from the Monterey Harbor in 1965. All four specimens (these data deposited in www.boldsystems.org) were identical to each other and to two others deposited in GenBank under the name *U. lobata*. The four sequences differed from those of *U. lactuca* and *U. rigida* by three bp, and *U. pertusa* by four bp. These data support the merging of *U. expansa* with *U. lobata*, a view implied by Hayden and Waaland (2004). Based on these results, however, it is not possible to pinpoint the time of the introduction of *U. pertusa* to Monterey.

—JEFFERY R. HUGHEY, Department of Science and Engineering, Hartnell College, Salinas, CA 93901. jhughey@hartnell.edu.

LITERATURE CITED

- ABBOTT, I. A. AND G. J. HOLLENBERG. 1976. Marine algae of California. Stanford University Press, Stanford, CA.
- AGUILAR-ROSAS, R., L. E. AGUILAR-ROSAS, AND S. SHIMADA. 2008. First record of *Ulva pertusa* Kjellman (Ulvales, Chlorophyta) in the Pacific coast of Mexico. *Algae* 23:201–207.
- DE CLERK, O., B. GAVIO, S. FREDERICQ, I. BARBARA, AND E. COPPEJANS. 2005. Systematics of *Grateloupia filicina* (Halymeniaceae, Rhodophyta), based on *rbcL* sequence analyses and morphological evidence, including the reinstatement of *G. minima* and the description of *G. capensis* sp. nov. *Journal of Phycology* 41:391–410.
- FREDERICQ, S. AND M. H. HOMMERSAND. 1989. Proposal of the Gracilariales ord. nov. (Rhodophyta) based on an analysis of the reproductive development of *Gracilaria verrucosa*. *Journal of Phycology* 25:213–227.
- FRESHWATER, D. W. AND J. RUENESS. 1994. Phylogenetic relationships of some European *Gelidium* (Gelidiales, Rhodophyta) species, based on *rbcL* nucleotide sequence analysis. *Phycologia* 33: 187–194.
- GUIRY, M. D. AND G. M. GUIRY. 2012. AlgaeBase. National University of Ireland, Galway. Website: <http://www.algaebase.org> [accessed 25 June 2012].
- HAYDEN, H. S. AND J. R. WAALAND. 2004. A molecular systematic study of *Ulva* (Ulvales, Ulvales) from the northeast Pacific. *Phycologia* 43:364–382.
- HEESCH, S., J. E. S. BROOM, K. F. NEILL, T. J. FARR, J. L. DALEN, AND W. A. NELSON. 2007. Genetic diversity and possible origins of New Zealand populations of *Ulva*. Biosecurity New Zealand Technical Paper no. 2007/01. Ministry of Agriculture and Forestry, Wellington, NZ.
- , ———, ———, ———, ———, AND ———. 2009. *Ulva*, *Umbraulva* and *Gemina*: genetic survey of New Zealand Ulvaceae reveals diversity and introduced species. *European Journal of Phycology* 44:143–154.
- HOLMES, E. M. 1896. New marine algae from Japan. *Journal of the Proceedings of the Linnean Society, Botany* 31:248–260.
- KAWAGUCHI, S., H. W. WANG, T. HORIGUCHI, G. SARTONI, AND M. MASUDA. 2001. A comparative study of the red alga *Grateloupia filicina* (Halymeniaceae) from the northwestern Pacific and Mediterranean with the description of *Grateloupia asiatica* sp. nov. *Journal of Phycology* 37:433–442.
- KIM, M. S., E. C. YANG, S. Y. KIM, I. K. HWANG, AND S. M. BOO. 2008. Reinstatement of *Gracilariopsis chorda* (Gracilariaceae, Rhodophyta) based on plastid *rbcL* and mitochondrial *cox1* sequences. *Algae* 23:209–217.
- , M. Y. YANG, AND G. Y. CHO. 2010. Applying DNA barcoding to Korean Gracilariaceae (Rhodophyta). *Cryptogamic Algology* 31:387–401.
- KIM, S. Y., F. WEINBERGER EINBERGER, AND S. M. BOO. 2010. Genetic data hint at a common donor region for invasive Atlantic and Pacific populations of *Gracilaria vermiculophylla* (Gracilariales, Rhodophyta). *Journal of Phycology* 46:1346–1349.
- KJELLMAN, F. R. 1897. Marina chlorophyceer från Japan. Bihang till Kongliga Svenska Vetenskaps-Akademiens Handlingar 23 (Afd. III, 11):1–44.
- KRAFT, L. G. K., G. T. KRAFT, AND R. F. WALLER. 2010. Investigations into southern Australian *Ulva* (Ulvophyceae, Chlorophyta) taxonomy and molecular phylogeny indicate both cosmopolitanism and endemic cryptic species. *Journal of Phycology* 46:1257–1277.
- LEE, J. I., H. G. KIM, P. L. J. GERALDINO, I. K. HWANG, AND S. M. BOO. 2009. Molecular classification of the genus *Grateloupia* (Halymeniaceae, Rhodophyta) in Korea. *Algae* 24:231–238.
- MILLER, K. A., L. E. AGUILAR-ROSAS, AND F. F. PEDROCHE. 2011. A review of non-native seaweeds from California, USA, and Baja California, Mexico. *Hidrobiológica* 21:365–379.
- OHMI, H. 1958. The species of *Gracilaria* and *Gracilariopsis* from Japan and adjacent waters. *Memoirs of the Faculty of Fisheries, Hokkaido University* 6:1–66.
- O'KELLY, C. J., A. KURIHARA, T. C. SHIPLEY, AND A. R. SHERWOOD. 2010. Molecular assessment of *Ulva* spp. (Ulvophyceae, Chlorophyta) in the Hawaiian Islands. *Journal of Phycology* 46: 728–735.
- VERLAQUE, M., T. BELSHER, AND J. M. DELOUS-PAOLI. 2002. Morphology and reproduction of Asiatic *Ulva pertusa* (Ulvales, Chlorophyta) in Thau Lagoon (France, Mediterranean Sea). *Cryptogamic Algology* 23:301–310.
- , P. M. BRANNOCK, T. KOMATSU, M. VILLALARD-BOHNSACK, AND M. MARSTON. 2005. The genus *Grateloupia* C. Agardh (Halymeniaceae, Rhodophyta) in the Thau Lagoon (France, Mediterranean): a case study of marine plurispecific introductions. *Phycologia* 44:477–496.
- YAMAMOTO, H. 1978. Systematic and anatomical study of the genus *Gracilaria* in Japan. *Memoirs of the Faculty of Fisheries, Hokkaido University* 25:97–152.